362-Pos Board B148
Computational Modeling of the Binding Mode of Transcription Activator-Like Effectors to DNA
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Transcription activator-like effectors (TALEs) form a novel class of DNA-binding proteins with predictable specificity. The TALE protein for *avrBs3* is composed of 17.5 repeat domains of 34 amino acid residues, and each repeat domain recognizes one specific base-pair. The specificity is encoded by the repeat-variable diresidue (RVD) of each repeat domain, and this modular nature of DNA base recognition enables useful applications in biotechnology. However, there is very little available structural information about the binding mode and the specificity. In this study, we attempt to generate possible binding modes of the TALE with *avrBs3* using protein structure modeling, protein-DNA docking, and molecular dynamics simulation. We discuss the observed binding specificities in terms of the molecular interactions found in the models.

363-Pos Board B149
Local Conformational Changes of Proteins in DNA Interfaces
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DNA-protein interaction plays a key role in many cellular functions such as transcription, replication, recombination and DNA packaging. Understanding the mechanisms by which the protein recognizes specific DNA sequences is one of the main topics in biology. With increased amount of crystal structures, conformational changes of proteins in DNA interfaces are suggested to be one of important factors in DNA recognition. To better understand a role of the deformation of the proteins, we carried out quantitative analyses of local conformational changes observed in DNA interfaces.
We first extracted crystal structures of DNA-unbound and DNA-bound states from PDB. We then described the backbone structures with the 7-letters-codes which were pre-defined for expressing conformations of 4 consecutive fragments. Using the letter codes, we identified conformational changes between DNA-unbound and bound states.
Then, we compared the amino acid composition and secondary structures of these conformationally variable regions in DNA interfaces with that in molecular surfaces. We found the regions in DNA interfaces tend to be more hydrophobic, less acidic residues, and have less secondary structure elements. We also found that conformations of the regions that bind to DNA in the minor groove are frequently changed and the width of the minor grooves is wider than that of the canonical B-DNA.

364-Pos Board B150
Insight into the Mechanism of Hu-Induced DNA Bending
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The structure and dynamics of the bacterial chromosome are maintained by many proteins. One of the most abundant proteins, a member of the DNABII family of DNA-binding proteins, is HU (Heat_Unstable). An architectural protein is a term used to describe proteins that introduce sharp bends (120-160 degrees) into DNA upon binding. DNA bending induced by these proteins is important for transcriptional regulation, initiation of replication, mu transposition, base excision repair, recombination, and negative supercoiling in bacteria. HU exhibits a strong preference for various distortions in DNA; nicks, gaps, cruciform, and sticky ends. The distorted DNA structures seem to share a similar binding motif of a flexible junction. There is little structural or dynamical information related to perturbation of these DNA motifs upon HU binding. A better understanding of HU binding to these motifs would provide insight into HU's role in the regulation of DNA structure. This information could also give insight into the general mode of recognition and binding for all non-sequence-specific DNA binding proteins found in both prokaryotes and eukaryotes. We have employed various fluorescence spectroscopy techniques to help answer questions related to this problem. Through the use of fluorescence anisotropy we have determined the dissociation constant in solution for HU binding to 3'overhang DNA substrate to be 3.41E-09+/- 1.2E-09 M, which is in good agreement with literature results. The stoichiometry for the specific binding of HU to these substrates is one to one, while non-specific binding may lead to higher order complexes. Fluorescence resonance energy transfer (FRET) results suggest bending of the DNA substrate upon HU binding. We hope to gain further information about the mechanism of HU-induced bending, structural recognition and binding through the use of such techniques as FRET mapping, Time-resolved fluorescence, and stop flow kinetics.

365-Pos Board B151
Characterization and Crystallization of BreR, the TetR Family Member Bile Response Repressor of Vibrio Cholerae
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How does *Vibrio cholerae* sense its environment and alter expression of its genes accordingly? Gaining a deeper understanding of the molecular basis of this process could lead to better strategies to prevent and treat cholera and other bacterial diseases.
My focus is on characterizing a bile response transcription regulator BreR and its interaction with DNA using X-ray crystallography and other biophysical techniques. BreR was expressed and purified. BreR formed a homodimer in solution and was homogeneous, according to analytical ultracentrifugation and gel filtration. Crystallization trials of BreR protein alone and the BreR-DNA complex are being carried out. The cocrystals of the BreR-DNA complex have been produced and a dataset at 3Å resolution has been collected.
The structure of BreR-DNA complex along with other biophysical characterization will give us information about how BreR interacts with DNA and about the ligand binding pocket of BreR. Eventually we hope this information would provide us a foundation to study the network of bile-regulated genes and hopefully help us be better able to prevent and treat cholera.

366-Pos Board B152
Multiple Binding Modes and DNA Organization by Non-Specific Binding of the *E. Coli* Nucleoid Associated Protein IHF
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The integration host factor (IHF) protein is an abundant E.Coli nucleoid protein. It is most well known as an essential cofactor in the process of phage Δ site-specific recombination where it binds to highly specific DNA sequences with a dissociation coefficient of less than 0.1 nM. Surprisingly, the intracellular concentration of IHF is in the range of 10 - 100 uM, extraordinarily larger than the need by its site-specific function. This raises an important question regarding the roles of non-specific binding of IHF that are yet to be understood. Here, we report several novel findings that IHF can bind to DNA with multiple distinct modes under physiological buffer conditions, and can organize DNA into highly condensed complex structures. Our findings have broad implications and highlight a role of IHF as an essential chromosomal DNA packaging protein.

367-Pos Board B153
Transition from Nonspecific to Specific DNA Recognition: Weak Frustration can Regulate Search Kinetics
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Rapid recognition of DNA target sites involves facilitated diffusion through which alternative sites are searched on genomic DNA. A key mechanism facilitating the localization of the target by a DNA-binding protein (DBP) is one-dimensional diffusion (sliding) in which electrostatic forces attract the protein to the DNA. As the protein reaches its target DNA site, it switches from purely electrostatic binding to a specific set of interactions with the DNA bases that also involves hydrogen bonding and van der Waals forces. High overlap between the DBP patches used for nonspecific and specific interactions with DNA may enable an immediate transition between the two binding modes following target site localization. By contrast, an imperfect overlap may result in greater frustration between the two potentially competing binding modes and consequently slower switching between them. A structural analysis of 125 DBPs indicates frustration between the two binding modes that results in a large difference between the orientations of the protein to the DNA when it slides compared to when it specifically interacts with DNA. Coarse-grained molecular dynamics simulations of in *silico* designed peptides that span the full range of frustrations between the two interfaces show slower transition from nonspecific to specific DNA binding as the degree of overlap between the patches involved in the two binding modes decreases. The complex search kinetics may regulate the search by eliminating trapping of the protein in semi specific sites while sliding.
Marcovitz, A., and Levy, Y. 2011. Frustration in protein-DNA binding imposes conformational switching and modulates search kinetics. *Proc. Natl. Acad. Sci. USA. In press*